Bioenergy

Today

aking the lead for the nation last year, the U.S. Department of Agriculture ordered all its locations nationwide to run vehicles and equipment on ethanol or biodiesel blends wherever feasible. Gasoline-operated vehicles and equipment will use a blend of at least 10 percent ethanol and 90 percent conventional gasoline. Diesel vehicles and equipment will use B20 biodiesel, a blend of 20 percent biodiesel and 80 percent regular diesel. The USDA fleet includes more than 700 "flex-fuel" vehicles, which use a blend of 85 percent ethanol and 15 percent gasoline.

The Department's action reflects the

KEITH WELLER (K9835-17)



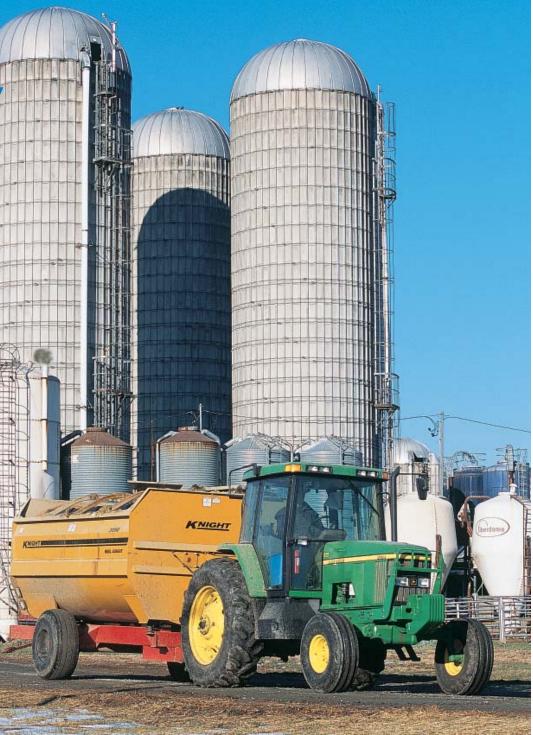
At the Beltsville Agricultural Research Center, quality assurance specialist David Johnson examines a sample of biodiesel while Greg Meyer, driver from a cooperating fuel company, fills a 20,000-gallon tank at a boiler plant, which heats BARC's dairy buildings.

federal government's commitment to expanding its use of biofuels and biobased products to set an example for the private sector. The year 2000 also saw a groundbreaking for a \$20 million National Ethanol Research Pilot Plant in Edwardsville, Illinois. When completed in January 2003, this will be the largest ethanol pilot plant in the country. The Agricultural Research Service is administering the federal government's contribution of \$14 million toward the \$20 million cost of construction. Industry considers the plant essential to meeting its goal of increasing annual ethanol production to 16 billion gallons over the next 10 to 15 years.

The new plant is designed to enable researchers to develop the technologies required to improve the efficiency of ethanol production. The more efficient the production, the lower the costs—in terms of both energy and money—and the more competitive ethanol becomes. Since each crop has its own mix of complex sugars and starches, each requires its own techniques to be economically processed into ethanol. The challenge is to design methods that allow different crops to be processed in the same facility.

Throughout the United States and in several other nations, scores of scientists are researching a wide variety of ways to improve ethanol production. These researchers include those in ARS' national Bioenergy and Energy Alternatives research program. At two of ARS' regional research centers—in Illinois and Pennsylvania—their goals are to improve the conversion of agricultural plant materials into ethanol and valuable coproducts, lower production costs and fuel emissions, and enhance performance properties of biodiesel. The Western Regional Research

Scientists worldwide are researching a wide variety of ways to improve ethanol and biodiesel production. The goals are to develop valuable coproducts, lower production costs and emissions, and enhance the performance of biofuels. ifferent



KEITH WELLER (K9842-10).



Above: In a biodieselpowered tractor, animal caretaker Angel Santiago heads to a dairy barn at BARC. The center uses B20, a common biodiesel blend, in its entire fleet of over 150 diesel vehicles.

Left: High-voltage electrician Alvin Coates fills the tank of an aerial hydraulic lift bucket truck with soy-based diesel.

Biofuel Basics

Biodiesel is a clean-burning alternative fuel that can be made from materials such as vegetable oils, animal fats, and spent cooking greases. Typically, biodiesel is prepared by the reaction of fat or oil with alcohol under alkaline conditions. Soybased biodiesel is the most commonly used form.

Ethanol is an alcohol-based fuel produced by fermenting sugars from crop starches. Currently, 95 percent of ethanol is produced from corn kernels. About 5 percent of U.S. ethanol is made from sugar- and starch-containing materials other than corn. These include wheat, barley, and sorghum grains; sugarcane; cheese whey; and wastes from paper mills, potato processing plants, breweries, and beverage manufacturers—or some combination of these materials.

Originally, most ethanol was made through wet-milling, which means the starch is separated from the corn germ and fiber and liquefied by cooking. The liquefying creates sugars in a form that can be fermented with yeast to produce ethanol and carbon dioxide. The ethanol is then removed from the slurry.

The number of ethanol plants has surged in the past few years, and drymilling is now the method used for over half of the ethanol currently produced. In this process, kernels are ground to a fine powder, and all of it is cooked to liquefy it, without removing the germ or fiber. Different enzymes are added at different stages and temperatures as the mash cools, producing ethanol and carbon dioxide.—By **Don Comis**, ARS.

Center, in California, focuses its biofuel efforts on ethanol. In 1999 the national program was expanded to include the breeding of improved energy crops.

Seeing Is Believing

USDA alone last year used over 100,000 gallons of biodiesel fuel and expects to easily double that amount next year. This puts practical examples of biofuel use within driving distance of

fleets to adopt biodiesel, particularly in Maryland. The cities of Greenbelt, Takoma Park, and Ocean City have all recently adopted biodiesel fuel for their snowplows and other public-works vehicles and equipment. Greenbelt also uses biodiesel to run its Connector bus, which ferries Greenbelters on short trips within the city, filling gaps in the Washington, D.C., metropolitan area public transit system. These cities

corn kernels for ethanol, such as *Amaiz*ing Oil, a new corn oil that can lower blood cholesterol levels, and a valuable food ingredient called *Zeagen*, a corn fiber gum. Both products were found in the fibrous hull that forms the kernel's outermost layer, and both are moving closer to the marketplace.

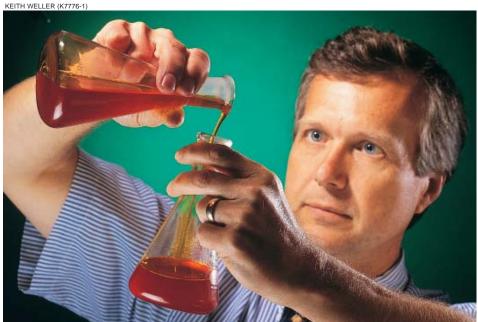
ERRC engineers have developed a radical alternative way to produce ethanol at a price expected to be significantly lower than is typical of conventional methods. It's called continuous fermentation with stripping. The method removes, or strips, ethanol contained in the escaping carbon dioxide, which is then recycled back to the fermentation vat. In the conventional process, when the ethanol level rises too high in fermentation vats, it slows the yeast's ability to produce more ethanol. The new method continuously strips ethanol from the fermentation broth, freeing the yeast to make additional ethanol.

The team has also developed a new process called pervaporation, which uses a membrane to filter ethanol out of the broth.

Says engineer Frank Taylor, "We are now looking for companies interested in taking our processes off the research bench and develop them further for commercial use."

Andy McAloon is the team's cost engineer. He developed a computer model that can estimate the cost per gallon of ethanol if a new process were used to produce it. "This guides our research so that we don't spend too much time on processes that would not yield a more competitive product," says Kevin Hicks, who leads the ERRC ethanol team. "It's expensive to test a process at the pilotplant stage, so this model could screen out processes not likely to be practical."

Other researchers at ERRC are working to reduce the cost of biodiesel production. They are making biodiesel fuel from lower quality starting materials, such as soybean soapstock (see story on page 9).



To help lower the cost of ethanol production, ARS scientists have developed valuable coproducts from corn, such as Amaizing Oil, which can lower blood cholesterol levels. Here, chemist Kevin Hicks checks the color and quality of a corn fiber oil sample.

public and private organizations around the country.

Leading by example, USDA's Henry A. Wallace Beltsville (Maryland) Agricultural Research Center (BARC) uses B20 biodiesel in its entire fleet of 150 diesel vehicles, including a tour bus (see cover photo) for the ARS National Visitor Center, located on BARC grounds. The center also heats some buildings with B5 (5 percent biodiesel).

The success with biodiesel in vehicles has encouraged commercial and public

learned about biodiesel by sending representatives to BARC meetings.

Cheaper Ways To Make Ethanol

Scientists at ARS' Eastern Regional Research Center (ERRC), in Wyndmoor, Pennsylvania, are working on lowering ethanol's price per gallon on two fronts: developing coproducts to defray costs and lower-cost production techniques and materials.

They've developed a growing number of valuable coproducts of processing

Enzymes for Greater Efficiency

At ARS' Western Regional Research Center, in Albany, California, scientists are creating better enzymes that produce ethanol in a more cost-effective manner.

"About 10 to 15 percent of the energy required to make ethanol goes toward providing the heat to cook the starch," says chemical engineer George Robertson. "The more energy it takes to make ethanol, the less useful it is as a fuel alternative. So we're working on enzymes that can digest the starch and make ethanol production more efficient. That could open up the ethanol market to other grains, like wheat," Robertson says.

To construct these enzymes, the team uses a technique developed in the pharmaceutical industry called directed evolution. Using biotechnology, they take apart key plant genes and reconstruct them, introducing mutations.

The mutant genes are then inserted into yeast organisms, where they begin to make, or express, starch-digesting enzymes. The scientists then screen the yeast colonies for their enzyme-producing abilities and select the best ones for another cycle of gene mutation and selection.

"We can do various things to direct the evolutionary path, speeding up development of enzymes with desired characteristics," says chemist Dominic Wong.

In the laboratory, at 98.6°F, their highpowered enzymes break down starch 50 times faster than the original enzymes. And the technique shows promise of making even better enzymes. The team plans to use similar approaches to develop new enzymes for use in biomass conversion.

Microbes, Too, Can Play a Role

Making biofuels, such as ethanol, economically from the whole crop instead of just the grain is the long-range goal of scientists in the Fermentation Biochemistry Research Unit at ARS' National

Center for Agricultural Utilization Research, in Peoria, Illinois.

"But our starting point is researching fermentation of fiber in just the corn kernel," says ARS microbiologist Rodney J. Bothast, who leads the project. Currently, the kernel fiber is separated out and used as inexpensive cattle feed that is valued for protein, not fiber. If technology were developed to break down the different polymers in kernel

component is not fermented but can be burned to produce energy.

"So far, the most effective way we've found to break down the fiber is to pretreat it with mild acid and then with alkaline hydrogen peroxide," says Bothast.

The pretreated fiber contains sugars, mainly arabinose and xylose and some glucose. Normally, ethanol-producing microbes eat the glucose first, leaving



Microbiologist Rodney Bothast (left) and technician Loren Iten add starter microorganisms to pilot-plant-size bioreactors in which ethanol is brewed from sugar mixtures derived from corn fiber.

fiber to simple sugars, about 10 percent more ethanol could be produced from each bushel of wet-milled corn.

Bothast collaborates with scientists at ERRC and in the Department of Wood Science at the University of British Columbia, Vancouver, in research on the physical and chemical pretreatment of fiber. Pretreatment frees the cellulose from hemicellulose, starch, and lignin components.

Cellulose fragments are more readily converted into sugars that can be fermented to make ethanol. The lignin little appetite for the other sugars. Nancy N. Nichols, microbiologist, and Bruce S. Dien, chemical engineer, have developed genetically engineered microorganisms that consume the sugars at nearly equal rates.

These researchers are collaborating with others at the University of British Columbia, Purdue University, and Williams Energy Service, in Pekin, Illinois—the second largest ethanol producer in the country—to test these new microbes on kernel fiber converted to sugars by industrial processes.

SCOTT BAUER (K9844-1)



Scientists are creating enzymes that produce ethanol in a more cost-effective manner. Technician Tina Williams and chemist Charles Lee use an automated liquid handler and a microplate reader to measure enzyme activity.

SCOTT BAUER (K9843-1)



At WRRC, technician Sarah Batt uses a robot to pick yeast colonies and transfer them onto starch plates, where they'll be screened for desirable enzyme production.

Value-Added Products

As the scientists seek ways to increase ethanol production efficiency, they're mindful of coproducts that might help make ethanol crops more economically successful. For example, other microbes developed by Nichols and Dien convert the sugars derived from kernel fiber into lactic acid. Biobased companies use lactic acid to produce solvents and biodegradable plastics.

Badal Saha, an ARS chemist, and microbiologist Timothy Leathers have developed yeasts that convert the xylose derived from corn fiber into xylitol, a low-calorie sweetener. Xylitol, which has a minty-cool taste, is used in some mints and gum and sells for about \$3 per pound. It's made from birch wood by an expensive, energy-intensive process.

Saha and Leathers have also discovered fungi that produce enzymes especially well suited for converting corn fiber into sugars. Use of enzymes decreases the amount of acid needed to convert corn fiber to sugars, and that makes ethanol an even more environmentally friendly fuel.

And Out on the Range . . .

Instead of making ethanol from the sugars and starches in plants, Ken Vogel, with ARS in Lincoln, Nebraska, is experimenting with using cellulose and hemicellulose from switchgrass as another source of ethanol. Vogel's hope is that farmers might be able to grow this native prairie grass on highly erodible soils —including those set aside for USDA's Conservation Reserve Program—harvest the grass periodically for ethanol production, and reap conservation benefits, such as reduced soil erosion and enhanced carbon storage.

Vogel and colleagues are breeding new switchgrasses for biofuel use. They're genetically improving the grass for conversion to ethanol and conducting on-farm trials to obtain economic information on production costs. Ron Follett, at Fort Collins, Colorado, is working with Vogel and ARS scientists at Mandan, North Dakota, to study carbon storage on lands grown for biofuel crops.

Plant geneticist JoAnn Lamb and colleagues at the ARS Plant Science Research Unit in St. Paul, Minnesota, are looking at alfalfa as another cellulose source for producing ethanol. They received \$288,000 from ARS' new \$2.4 million in funding for developing bioenergy crops.

They are breeding a new alfalfa variety specifically to double as a high-quality livestock feed and a bioenergy crop. They'll incorporate genes from southern European varieties to give the plant a thicker, almost woody, stem. This means more cellulose for ethanol production.

The humid East might prefer alfalfa to switchgrass as an ethanol source, but switchgrass is ideally suited for the arid West, because it needs very little rainfall to grow. Both alfalfa and switchgrass can also be burned to generate electricity.

There are obstacles to overcome when making ethanol from cellulose in plants like switchgrass or alfalfa, such as finding ways to convert the complex sugars in cellulose into simple ones that can be fermented to produce ethanol. Facilities to do this conversion will have to be built. Equipment for this purpose could be tested at the new Illinois pilot ethanol plant when it is up and running, as could equipment for the new continuous fermentation stripping process.—By Don Comis, ARS. Ben Hardin and Kathryn Barry Stelljes, both formerly with ARS, also contributed to this story.

This research is part of Quality and Utilization of Agricultural Products (#306) and Bioenergy and Energy Alternatives (#307), two ARS National Programs described on the World Wide Web at http://www.nps.ars.usda.gov.

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